REMARKS

With entry of the present amendment, claims 1, 2, 6 – 18, 21 – 34 and 37 – 51 are pending. Claims 3 – 5, 19, 20, 35, 36 and 52 have been cancelled. Claims 1, 6, 10, 14, 17, 21, 22, 33 and 46 – 51 have been amended. Claims 1, 17, 33, 43, 44, and 45 are independent claims. Claims 1, 17 and 33 have been amended to include the phrase "without adjusting the pH of the slurry or solution of the starch". The dependency of claims 6, 10, 14, 21, 22 and 46 has been changed. Additionally, claims 46 – 51 have been amended to include that the liquefact is characterized as having a pH of about 4.0 to 4.5 and is suitable for saccharification without further adjustment of the pH. New matter has not been introduced by the instant amendment.

The claims in the instant application have been rejected under 35 U.S.C. §102 and §103. There are no other pending rejections. With respect to the §102 and §103 rejections, two references have been cited. The primary reference is Shetty et al. "Factors Affecting the Economics of Glucose Production", Delivering Innovation Through Biotechnology", Genencor International, Inc. (1998) (Shetty). Shetty is also a co-inventor of the instant invention. The secondary reference is JP 10-136979.

The Shetty reference reviews the process for starch liquefaction and highlights areas that could be improved to reduce the economic costs involved in this process. Specifically highlighted is the desire to perform liquefactions at a lower pH which would decrease the chemical demand for pH adjustments prior to and after liquefaction, reduce color and by-product formation and lower refining requirements. As taught in the reference, a number of alpha amylases used in the liquefaction process require a pH greater than 6.0. However, pHs greater than 6.0 produce maltulose which is an undesirable by-product of the liquefaction process. Moreover, after liquefaction a pH adjustment is used prior to glucoamylase saccharification, which occurs at pH 4.2 to 4.5.

The reference reveals that one advance in the liquefaction process would be use of an alpha amylase with a lower pH tolerance. As taught by the reference, an

alpha amylase having a pH tolerance, less than pH 6.0 would potentially produce less maltulose, decrease the reaction rate and increase glucose yield.

The reference further describes one solution to this problem, and this is the use of a low pH thermostable alpha amylase from *B. licheniformis.* This alpha amylase designated GC521 (see page 12) performs at a pH of 5.6 and produces a starch hydrolzyate composition, which is identical to that of alpha amylases previously used in the industry (i.e. SPEZYME AA).

There is no discussion in the Shetty reference concerning an alpha amylase from *Bacillus acidocaldarius.* Additionally, there is no teaching or suggestion in the reference concerning a process for starch liquefaction wherein an alpha amylase having a pH stability below 5.0 could be used to obtain a starch liquefact having a DE of about 10 – 12 in 60 to 75 minutes after adding the alpha amylase. Nor does the reference teach or disclose a maltulose free starch liquefact characterized as comprising an acid stable alpha amylase obtained from *Bacillus acidocaldarius* wherein the amylase has been optionally thermally inactivated and wherein the liquefact has a pH 4.0 to 4.5 and is suitable for saccharification without pH adjustment.

The process disclosed at page 6 of Shetty teaches that a starch slurry is generally adjusted to pH 6.0 to 6.5, a thermostable acid alpha amylase is then added, temperature is increased to about 105 °C to produce a soluble dextrin solution which is discharged and cooled to about 95°C and then the size of the dextrin is further reduced by the alpha amylase until a final level of 10 – 12 DE.

Page 14 of Shetty discloses the improvement in using GC521. Figure 1 represents pH stability. With respect to low pH stability of GC521, after 60 minutes a DE around 4 is obtained at pH 5.0 – 5.3 and a DE of about 7 is obtained at pH 5.6 to 5.9 at 95C. Figure 2 depicts calcium independence at pH 5.6 and figure 3 depicts temperature stability at pH 5.6. There is no teaching concerning an alpha amylase from a strain of *Bacillus acidocaldarius* and further there is no teaching or suggestion of an alpha amylase, which is able to produce a starch liquefact with a DE of 10 – 12 in 60 to 75 minutes at a pH of less than 5.0.

As stated repeatedly in the present disclosure, the liquefaction step does not require pH adjustment and further less time is required to produce a liquefact with a

commercially acceptable DE value (see page 4, lines 13 – 16 and page 5, lines 13 - 19).

JP Appln. No. 08302245 discloses the *Bacillus acidocaldarius* strain, KSTM-2037 which is stable in the pH range of 4.5 to 5.0 under heating at 90°C for 15 min and having a pH optimum around 4.0. The enzyme is disclosed to be stable up to 80°C, when it is kept at pH 4.5 for 15 minutes and has an optimal temperature of 80 – 90°C.

Applicants assert not only is the process patentable over the cited art but also the claimed starch liquefact product is novel and unobvious over the cited art.

Rejection under 35 U.S.C. §102(b)/103 -

Product by process claims 46-52 were rejected under 35 U.S.C. §102 as being anticipated by or, in the alternative under 35 U.S.C. §103 (a) as obvious over Shetty.

While the product taught in Shetty may have a DE that is the same as the DE of the claimed liquefact, the claimed liquefied starch product includes an alpha amylase obtained from a *Bacillus acidocaldarius* and not *Bacillus licheniformis*. Moreover, the claimed liquefact has a pH that is in the range of pH 4.0 to 4.5 and is suitable for saccharification without further pH adjustment. The alpha amylase enzyme that comprises the claimed liquefact may or may not be a thermally inactivated enzyme.

The Examiner states at page 3 of the office action dated March 24,

> "It is noted that the enzyme used to produce the claimed product is from a different species of microorganism than the prior art enzyme. However, even if this results in a **nominal difference** between the reference product and the claimed product such that there is, in fact no, anticipation, the reference product would, nevertheless, have rendered the claimed product obvious to one of ordinary skill in the art.... Thus the claimed invention as a whole was clearly *prima facie* obvious .... "

Applicants contend the reference, which teaches liquefact comprising an alpha amylase obtained from *Bacillus licheniformis*, clearly does not anticipate the

claimed liquefact product comprising an alpha amylase from *Bacillus acidocaldarius*, and further Applicants contend the Examiner has not meet the burden of proving a *prima facie* case of obviousness. While the Shetty reference suggested it would be beneficial to the industry to use an alpha-amylase having a lower pH than the conventional thermostable alpha-amylases used in the industry at the time, the enzyme that is taught is a thermostable alpha amylase obtained from *B. licheniformis* which is taught to operate at a pH greater than 5.0. There is no suggestion or any expectation of success provided by the reference directed to a starch liquefact characterized as having a pH of about 4.0 to 4.5 which is free of maltulose and suitable for saccharification without adjustment of the about 4.0 – 4.5 pH.

The difference between the liquefact and process as disclosed in Shetty and the claimed liquefact (and the claimed process as discussed below) is not a nominal difference but a significant difference and provides potential improvement beyond the teachings of Shetty.

Rejection under 35 U.S.C. §103 -

The Examiner has maintained the rejection of claims 1 – 52 as being unpatentable over Shetty in view of JP10-136979.

Applicants incorporate the argument presented above concerning Shetty. The Examiner has stated,

> "Shetty discloses a process of preparing glucose from starch said process using the claimed process parameters. See, e.g. pages 6 and 14. Shetty differs from the claims in that Shetty uses a different alpha-amylase enzyme than that recited in the claims. However, Shetty disclosed that alpha amylase active at acidic pH are advantageous in processes of producing glucose from starch..."

Applicants disagree with the Examiner's statement not only does Shetty use a different alpha amylase which is clearly not taught or suggested in the reference, but also the claimed process parameters are not identical or reasonably suggested. While Shetty may disclose parameters that could be modified in the liquefaction process which would enhance the efficiency of the commercial process and further

discloses a particular amylase (GC521 obtained from *B. licheniformis*) this enzyme is not disclosed as performing at a pH of 4.0 to 4.5 nor of obtaining a DE of 10 – 12 within 60 – 75 minutes of adding the alpha amylase. In order to establish a *prima facie* case of obviousness the reference must provide sufficient basis for the required expectation of success.

Recognizing one of the deficiencies of Shetty, the Examiner has cited JP10-136979 as teaching the *Bacillus acidocaldarius* (KTSM #2037) which is included in the instant claims. However, it is submitted that JP 10-136979 in combination with Shetty does not suggest to one skilled in the art that alpha amylase obtained from *Bacillus acidocaldarius* (KTSM #2037) could be used in a liquefaction process wherein a DE of about 10 – 12 is obtained within 60 to 75 minutes of adding the alpha amylase nor that the pH of the starch solution would not have to be adjusted prior to or after the addition of the alpha amylase to be useful for saccharification.

Applicants submit even if a *prima facie* case of obviousness were established, Example 1 of the present application demonstrates unexpected results. In this example, *Bacillus licheniformis* derived alpha amylases (an amylase sold under the brand name SPEZYME FRED L), a *Bacillus stearotheromphilus* derived alpha amylase (an amylase sold under the brand name TERMAMYL SC), and the *Bacillus acidocaldarius* (KTSM #2037) are used under the same environmental conditions. Only the use of the *B. acidocaldarius* provided the desired DE level within 60 to 75 minutes after application. Reference is made to page 12, lines 24-30 and Fig. 4.

Additionally, JP-10-136979 describes KSTM 2037 as stable for 15 minutes in the pH range of 4.5 to 5.0 under heating at 90°C for 15 minutes with an optimal temperature between 80-90° C. An element of Applicants' claims include heating at a temperature of at least 90°C and in some case heating at a temperature of 90 – 155°C (claim 1), 105 – 110°C (claim 43), and 140 – 155°C (claim 44).

Applicants respectfully request that the rejections based on 35 U.S.C. §102 and 35 U.S.C. §103 be withdrawn. Claims 1, 2, 6 – 18, 21 – 34 and 37 – 51 are in condition for allowance and allowance of said claims is kindly solicited.

Respectfully submitted,

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